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Culture-independent detection of *Campylobacter* by metagenomic sequencing of faecal samples

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Our task

- To use open-source software to detect *Campylobacter* in metagenomic datasets from sequencing of artificially inoculated faecal samples from chicken and humans
- To define a detection limit using this method
- To look at diversity among identical samples spiked with different levels of *Campylobacter*

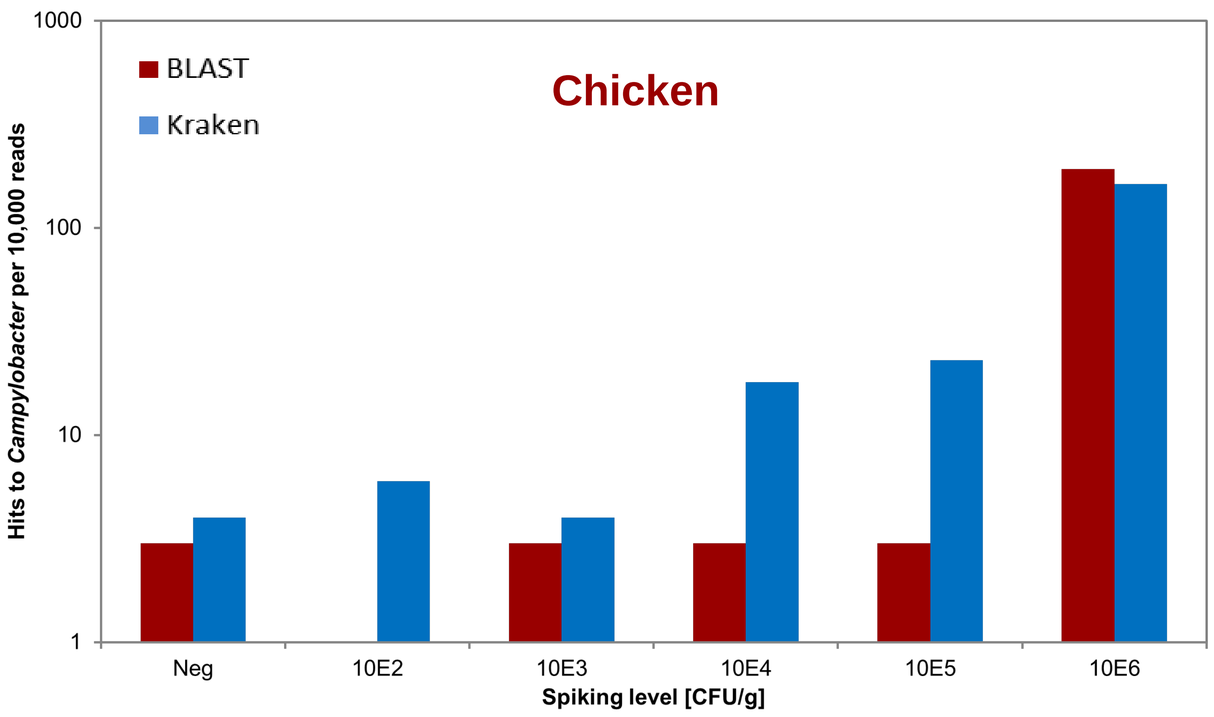
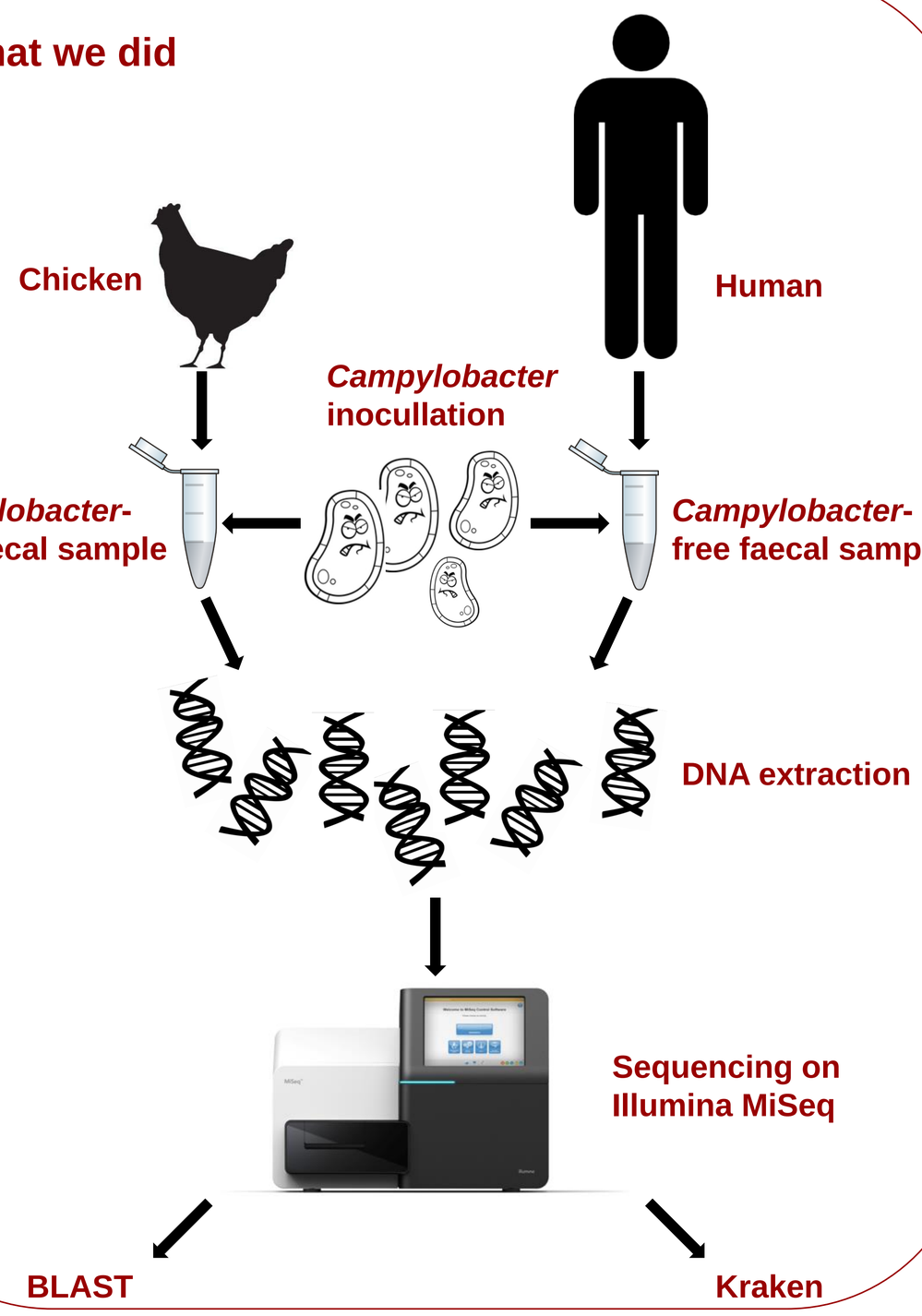


Figure 1 – Hits to *Campylobacter* found using BLAST (red) and Kraken (blue)
Detection of *Campylobacter* in chicken faecal samples is possible from 10^6 CFU/g using BLAST (red bars) and from 10^4 CFU/g using Kraken (blue bars). For BLAST results hits are number of contigs matching *Campylobacter* in proportion to the total number of contigs. For Kraken results hits are number of reads assigned to *Campylobacter* in proportion to the total number of reads.

What we did



Our conclusion

Detection limit	BLAST	Kraken
Chicken	10^6	10^4
Human	10^7	10^6

- High detection limits – 10^4-10^7 CFU/g
- Detection limits are lower in chicken faecal samples than in human faecal samples
- Kraken is slightly better at detection than BLAST
- Chicken faecal samples derived from same matrix and spiked with different levels of *Campylobacter* are 84-99% similar and the most abundant genera are *Lactobacillus*, *Escherichia*, and *Bacteroides*

Figure 2 – Hits to *Campylobacter* found using BLAST (red) and Kraken (blue)
Detection of *Campylobacter* in human faecal samples is possible from 10^7 CFU/g using BLAST (red bars) and from 10^6 CFU/g using Kraken (blue bars). For BLAST results hits are number of contigs matching *Campylobacter* in proportion to the total number of contigs. For Kraken results hits are number of reads assigned to *Campylobacter* in proportion to the total number of reads.

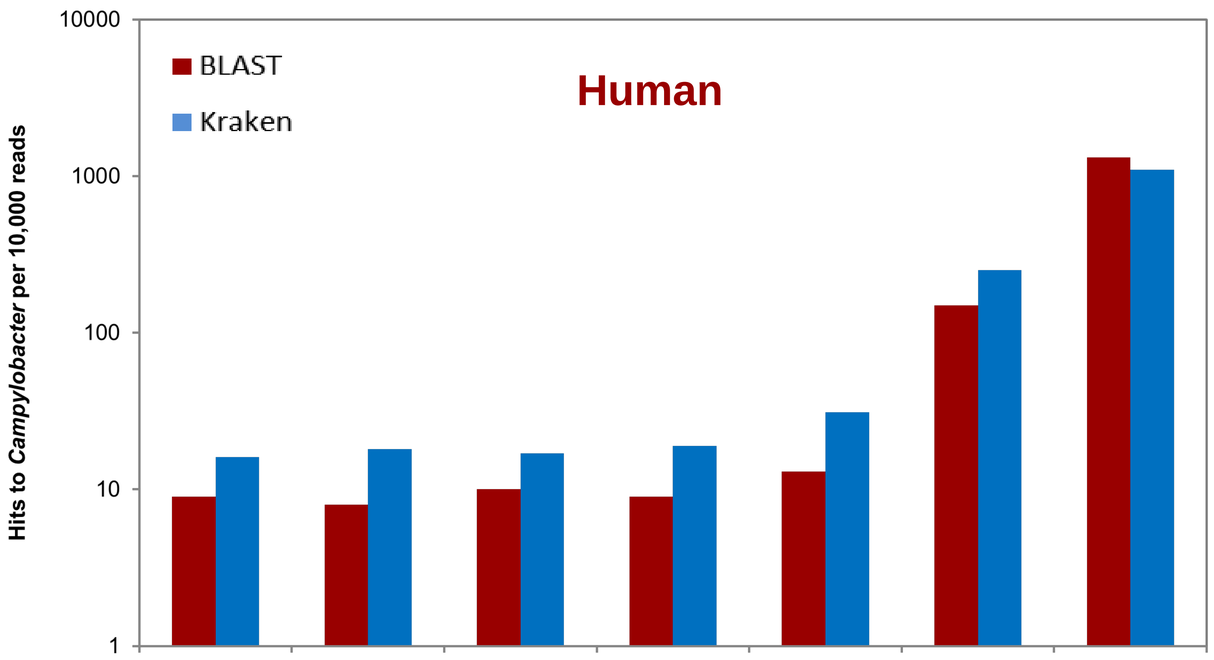
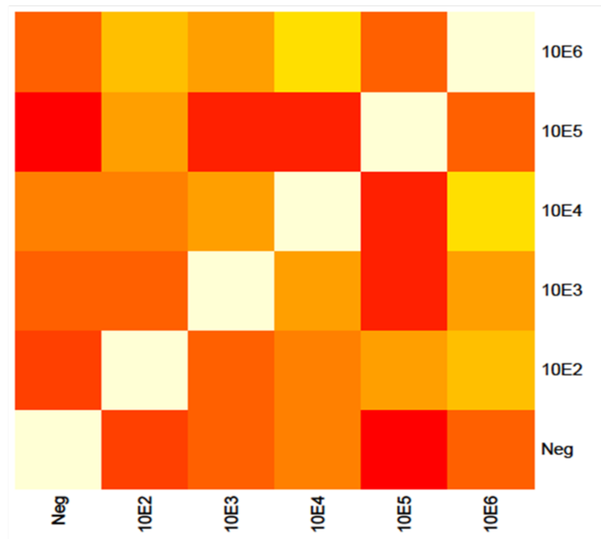


Figure 3 – Similarity in diversity
84-99% similarity is seen in diversity among the chicken faecal samples. We observe a lower similarity in the faecal sample composition than expected, since they derive from the same faecal matrix. We speculate that this is due to heterogeneity within the matrix.



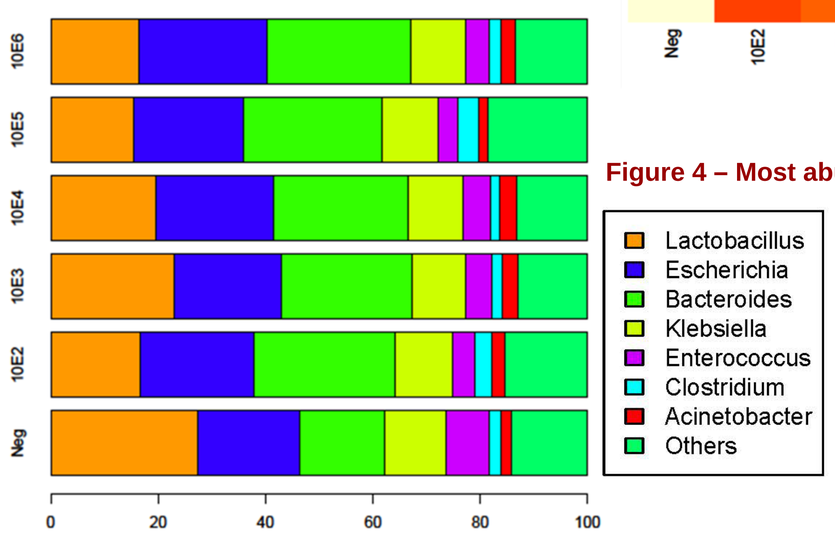


Figure 4 – Most abundant genera
Most abundant genera in the chicken faecal samples are *Lactobacillus*, *Escherichia*, and *Bacteroides*. There are more variation among the samples than we expected as they derive from same faecal matrix. Even the most abundant genus is not the same in all samples.

Next steps

- Try other software programs for detection
- Find a software solution for typing
- Look for other pathogenic bacteria
- End goal: To replace culturing and molecular analyses by diagnostic metagenomics used for detection and typing in surveillance and outbreak investigation

References
Hoorfar, J.; Rapid Detection, Characterization and Enumeration of Foodborne Pathogens; 2011